METHOD AND APPARATUS FOR MAKING MAGNETICALLY RESPONSIVE NANOPARTICLES

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority of U.S. Provisional Patent Application Serial No.: 60/479,381 filed June 18, 2003.

FIELD OF THE INVENTION

[0002] The present invention relates generally to the production of nanospheres, and more particularly, to nanospheres having single domain magnetically responsive nanoparticles.

SUMMARY OF THE INVENTION

[0003] The present invention is directed to a nanosphere having a diameter less than 300 nanometers. The nanosphere comprises at least one magnetically responsive nanoparticle having single domain properties and a bio-compatible shell encapsulating the nanoparticle.

[0004] The invention further includes a nanosphere having a diameter less than 300 nanometers. The nanosphere comprises at least one magnetically responsive nanoparticle and a bio-compatible shell encapsulating the nanoparticle. The nanoparticle is prepared by a process comprising vaporizing a magnetic metal salt, oxidizing the vaporized magnetic metal salt to produce an oxidized metal vapor, and quenching the oxidized metal vapor.

[0005] Still further, the present invention comprises a nanosphere having a diameter of less than 300 nanometers. The nanosphere comprises at least one magnetically responsive nanoparticle and a bio-compatible shell encapsulating the nanoparticle. The nanoparticle has single domain properties and is prepared by a process comprising preparing a solution comprising a mixture of magnetic metal salts and alkaline media to form a precipitate, washing

the precipitate with a solvent, magnetically collecting the precipitate, washing the precipitate again with the solvent, and drying the precipitate.

[0006] Still yet, the present invention includes a method for making a magnetically responsive nanoparticle. The method comprises vaporizing a magnetic-metal salt, oxidizing the vaporized magnetic-metal salt to produce an metal oxide vapor, and quenching the metal oxide vapor to produce at least one nanoparticle of a desired diameter.

[0007] The present invention further includes a method for making a magnetically responsive nanoparticle. The method comprises forming a precipitate by mixing a magnetic-metal salt and an alkaline media, collecting the precipitate using a magnetic field, and drying the precipitate.

[0008] Further still, the present invention includes a magnetically responsive nanosphere having a bio-compatible shell. The nanosphere is prepared by a process comprising atomizing a nanodispersion and drying the atomized nanodispersion in a magnetic field. The nanodispersion comprises a magnetically responsive nanoparticle and sodium silicate.

[0009] The present invention further comprises a magnetically responsive nanosphere having a bio-compatible shell. The nanosphere is prepared by a process comprising atomizing a dilute solution to form at least one droplet and drying the droplet in a magnetic field to remove the solvating media. The dilute solution comprises at least one magnetically responsive nanoparticle, a solvating media, and a bioerodable polymeric material.

[0010] The present invention further includes a nanosphere having a diameter of less than 300 nanometers. The nanosphere comprises a plurality of single domain superparamagnetic magnetite nanoparticles having uniformly aligned magnetic moments. A collagen shell encapsulates each of the plurality of nanoparticles. An outer bio-compatible shell encapsulating the nanoparticles.

BRIEF DESCRIPTION OF THE DRAWINGS

[0011] Figure 1 is a diagrammatic representation of a nanosphere delivery system constructed in accordance with the present invention. The nanosphere has a plurality of nanoparticles and a therapeutic surrounded by a bio-compatible shell.

[0012] Figure 2 is a diagrammatic representation of an alternative embodiment of a nanosphere delivery system constructed in accordance with the present invention. The nanosphere of Figure 2 comprises a plurality of nanoparticles surrounded by a bio-compatible therapeutic.

[0013] Figure 3 is a diagrammatic representation of an alternative embodiment of a nanosphere delivery system constructed in accordance with the present invention. The nanosphere of Figure 3 comprises a plurality of magnetically responsive nanoparticles encapsulated within a bio-stable silica shell. The nanoparticles are positioned so that they have uniformly aligned magnetic moments.

[0014] Figure 4 is a diagrammatic representation of a gas phase synthesis system for producing magnetically responsive nanospheres using an rf inductive plasma torch.

[0015] Figure 5 is a diagrammatic representation of an rf-IP torch used in a process to make nanoparticles in accordance with the present invention.

[0016] Figure 6 is a diagrammatic representation of a system used to produce nanospheres containing single domain superparamagnetic nanoparticles having uniformly aligned magnetic moments.

DESCRIPTION OF THE PREFERRED EMBODIMENT

[0017] Targeted delivery of therapeutics to a specific site within a body provides advantages over oral or systemic administration. For example, effective doses of therapeutics may be delivered at lower amounts to a desired target without exposing the entire body to adverse conditions or side effects. Drug delivery systems based on magnetically responsive nanoparticles provide a method for external control and site-specific delivery of therapeutics.

[0018] The present invention is directed to processes and methods for making nanospheres comprising single domain nanoparticles. Further, the present invention is directed to the structure of nanospheres comprised of magnetically responsive nanoparticles.

[0019] Turning now to the drawings in general and Figures 1-3, in particular, there is shown therein a representation of a nanosphere 10a-c in accordance with the present invention. The nanosphere of Figure 1 comprises at least a magnetically responsive nanoparticle 12 and a bio-compatible shell 14a encapsulating the nanoparticle. Figure 1 illustrates the usefulness of nanospheres having magnetically responsive nanoparticles by demonstrating that a therapeutic 16 may be encapsulated within the bio-compatible shell 14a. The combination of magnetically responsive nanoparticles 12 and therapeutics 16 encapsulated within a bio-compatible shell 14a provides a system that may be delivered to a specific target within an organism using magnetic vectoring.

[0020] Continuing with Figure 1, there is shown a nanosphere 10a, prepared using a method described herein. The nanosphere 10a of Figure 1 comprises a plurality of magnetically responsive nanoparticles 12 in an erodable polymer matrix (not shown) and encapsulated within the bio-compatible shell 14a. The nanosphere 10a of Figure 1 contains the therapeutic 16, which is further encapsulated within the bio-compatible shell 14a. The nanosphere 10a generally has a diameter of less than 300 nanometers, and more preferably a diameter of 100 nanometers or less.

[0021] The bio-compatible shell 14a of nanosphere 10a may comprise materials, such as collagen, albumin, and polylactic acid, that are capable of being internalized by a cell. The bio-

compatible shell 14a encapsulates the nanoparticles 12 and forms a reservoir within which the therapeutic 16 may be contained. Other natural polymers, or synthetic bioerodable polymers, for example, polylactides or polyglycolides, or other similar materials known to those skilled in the art may also be used.

[0022] The bio-compatible shell 14a may further comprise an outer surface 18 that has cell adhesion factors 20 supported on the outer surface 18 of the bio-compatible shell. The use of cell adhesion factors allows the production of nanospheres that have a special affinity for a target cell. Thus, the cell adhesion factor 20 may comprise a protein having an affinity for a predetermined type of cell. It will be appreciated that a wide array of cell adhesion factors may be used with nanospheres of the present invention without departing from the spirit of the invention.

[0023] As shown in Table I, various adhesion factors can be used to enhance cell endocytosis, that is, to facilitate engulfing of the drug encapsulated in the nanospheres 10a and 10b by the target cell.

Table I

Adhesion Factors	**Target cells for adhesion**
Collagen Type I	Epithelial cells Muscle cells Nerve cells
Collagen Type II	Chondrocytes
Collagen Type IV	Epithelial cells Enodothelial cells Muscle cells Nerve cells
Superfibronectin	Epithelial cells Mesenchymal cells Neuronal cells Fibroblasts Neural crest cells Endothelial cells
Victronectin	Platelets Endothelial cells Melanoma cells

	Osteosarcoma
Selectins	Endothelial Cells Platelets Leucocytes

[0024] Continuing with Figure 1, the bio-compatible shell 14a may encapsulate an erodable polymer matrix (not shown) that entraps the therapeutic 16 and releases it at a rate dependent upon the rate at which the matrix dissolves. It is preferable that the erodable polymer matrix is non-toxic and capable of being consumed, metabolized or expelled by the cell. An example of such an erodable polymer matrix is collagen, or any other suitable natural or synthetic polymer. In some instances with therapeutic delivery applications (discussed hereinafter), it may be desirable to form the nanosphere without the erodable polymer matrix, producing a nanosphere including a magnetically responsive nanoparticles 12 with a bio-functional component, or a therapeutic 16, as the encapsulating material. The physical properties of the therapeutic 16 have no relative effect on the functioning of the delivery system, because the delivery mechanism is externally controlled. The therapeutic 16 is delivered to the desired site, independent of its physical chemical properties, thus it can be water soluble or insoluble. Once internalized by the cell, the therapeutic 16 is exposed to the cellular components and consumed. The erodable polymer matrix serves to control the rate of release of therapeutic 16 from the nanosphere 10a. A tightly cross-linked matrix will exhibit a slow release rate providing low doses over longer periods of time. When no erodable matrix is present a rapid release of therapeutic 16 can be expected.

[0025] As previously discussed, nanosphere 10a of Figure 1 comprises at least a magnetically responsive nanoparticle 12 having single domain properties. However, it will be appreciated that nanosphere 10a may comprise a plurality of magnetically responsive nanoparticles 12. Preferably, the nanoparticles 12 are situated such that the single domain magnetically responsive nanoparticles have uniformly aligned magnetic moments. The nanoparticles 12 may be comprised of a ferrite such as magnetite and have a silica or titania

coating 22. Use of a such a coating 22 on the nanoparticle 12 renders the nanoparticle biocompatible.

[0026] Magnetite nanoparticles 12 are highly active ferromagnetic materials and are superparamagnetic, being magnetic when in a magnetic field and losing this property when the field is removed. The single domain properties of the magnetite nanoparticles 12 of the present invention, when in a magnetic field, will only be attracted to the strongest side of the field gradient and will not be attracted by other or similar nanoparticles. Thus, particle to particle interactions resulting in clumping or other undesirable effects are minimized. Once the magnetic field is removed, the nanoparticles 12 lose their magnetic remanence.

[0027] Turning now to Figure 2, there is shown therein an alternative nanosphere 10b having a bio-compatible shell 14b comprising a biostable coating that makes the bio-compatible shell biostable. Nanosphere 10b is shown to contain a plurality of nanoparticles 12 within the bio-compatible shell 14b. The nanoparticles 12 are arranged so that the magnetic moments of each are aligned with the other nanoparticles. The bio-compatible shell 14b may have cell adhesion factors 20 as previously discussed. The use of nanospheres 10b comprising a bio-stable shell 14b promotes sustained residence of the nanoparticles 12 within targeted cells as discussed hereinafter.

[0028] Turning now to Figure 3, there is shown therein a nanosphere 10c comprising a plurality of single domain nanoparticles 12 encapsulated by a biocompatible shell 14c. However, the nanosphere 10c of the Figure 3 is formed so that the therapeutic to be delivered to the cell forms the biocompatible shell 14c. The therapeutic 16 may be coupled or physically attached to the nanoparticles 12 by chemical means that will be apparent to one skilled in the art. In some instances, for example, as in an application for chemotherapeutic delivery, linkage of the therapeutic 16 to the nanoparticle 12 surface may be necessary to "drag" the therapeutic magnetically to the site. Such a linkage may be created by adding such compounds as linkers or

functional groups to the silica surface 22 of the nanoparticle 12 so that the surface coating comprises "hooks" (not shown) by which the therapeutic 16 may be linked to the nanoparticles. "Hooks" is to be understood as a generic term to denote a physical attribute, affinity site, functional moiety or mechanism by which the therapeutic 16 may be linked. The hooks can be, for example, physical locations at which the therapeutic may be physically or chemically attached.

[0029] Turning to Figure 4, there is shown therein a system for preparing magnetically responsive nanospheres 10a-c having magnetically responsive nanoparticles 12 and bio-compatible shells 14a-c. The magnetically responsive nanoparticle 12 (Figures. 1-3) is prepared by a plasma synthesis process comprises vaporizing a magnetic metal salt, oxidizing the vaporized magnetic metal salt, and quenching an oxidized metal vapor produced in the oxidizing step.

[0030] Figure 4 shows a diagrammatic representation of a radio-frequency-inductive plasma (rf-IP) synthesis system, based on an electrodeless system, to prepare magnetically responsive nanoparticles 12. The magnetic metal salt is heated so that the magnetic metal salt is vaporized. As an effective heat source, plasmas can generate temperatures above 10,000 °K, far above the melting temperatures of known materials. It is to be understood, that other heat sources known to those skilled in the art, such as, for example, gas burners, may be used. rf-IP, however, allows a relatively large volume throughput versus low velocity plasma gas over range of reactor conditions of pressure and temperature. As a result, nanoparticle size and distribution can be precisely controlled.

[0031] Once the magnetic metal salt is vaporized it may oxidized. The preferred plasma synthesis process for making magnetite-based nanoparticles involves the vaporization and injection of the magnetic metal salt in the presence of oxygen in an rf-IP torch 32 from direction 34. A shown in Figure 5, the rf-IP torch 32 may comprise a ceramic shell 36 and an

induction coil 38. The base 40 of the plasma torch 32 is connected to a reactor 42. The magnetic metal salt may comprise ferric and ferrous mixture having a ratio between 2 to 1 and 10 to 1. The magnetic metal salt may further comprise a ferric salt or ferric/ferrous salt combination (3:1), for example chloride.

[0032] Referring now to Figure 5, the magnetic metal salt mixture may be injected into the plasma reactor 32 via an opening 44. The magnetic metal salt is vaporized in the presence of oxygen, which is injected into the torch via a gas inlet 46. The vaporized magnetic metal salt feed may be axially injected into the center of the plasma discharge 47, or it could be injected in the radial direction into the plasma discharge 47 at the exit of the torch, or a combination of the two modes of injection could be used. Subsequent to the injection, the vaporized magnetic metal salt feed reacts with oxygen in the plasma where oxidation of the magnetic metal salt occurs to produce an oxidized metal vapor. The following oxidation reaction proceeds rapidly to yield the formation of, for example, Fe_3O_4 vapors and free chlorine:

$$6FeCl_3 + 2O_2 \rightarrow 2Fe_3O_4 + 9Cl_2.$$

[0033] Salts, such as, for example, Li^+ may be additionally injected in the reactor to create surface charges to reduce collisions and minimize particle agglomeration. Additionally, if desired, the nanoparticles 12 may be treated with a bio-compatible surface agent. Surface treatment agents such as silicon tetrachloride or titanium tetrachloride can be introduced immediately downstream in the reactor to cause the ferrite nanoparticles to have Si or Ti coatings respectively. The silicon tetrachloride or titanium tetrachloride may be injected simultaneously with the magnetic metal salt into the reaction gas stream in the rf-IP torch chamber via an optional inlet, or downstream from reaction gas stream, or a combination of simultaneously with and downstream from the reaction gas stream. The formed vapors in the chamber co-condense giving rise to a spherical shell possessing a magnetically responsive nanoparticle with a surface

layer of titania or silica, and therefore, result in formation of nanoparticles that are biocompatible.

[0034] It is to be understood that titanium tetrachloride and silicon tetrachloride are only representative examples of materials used for biocompatibility and coating, are not limited to those materials. Rather, other materials will be apparent to one skilled in the art. Further, suitable organic monomers and polymers may also be used to coat the magnetically responsive nanoparticles.

[0035] Returning to Figure 4, the oxidized metal vapor that has formed in the reactor 42 is subjected to controlled quenching by passing the reactor stream into a quench chamber 48. In the quench chamber 48, rapid gas expansion occurs concurrently with the injection of an inert cooling gases to yield nanoparticles of uniform size and size distribution. Quenching of the nanoparticles may be achieved by injection of a compressed gas, for example air, that creates a quench zone which rapidly reduces the temperature of the particles, thus effectively terminating particle growth to yield uniform particle size and size distribution. Controlled quenching enables formation of a relatively narrow particle size distribution centered around a target mean particle diameter of, for example, less than 30 nanometers, preferably less than 10 nanometers. The nanoparticles may then be collected using on electrostatic filters or similar type systems known to those skilled in the art.

[0036] In another exemplary embodiment, magnetically responsive nanospheres having a single domain nanoparticle and bio-compatible shell can be prepared by a generally aqueous process. Generally known methods for aqueous synthesis may be modified to prepare the nanoparticles for the purpose of this invention. For example, the method disclosed in Massart (IEEE Transactions on Magnetics, col. Mag-17, No. 2, 1247 March 1981, the contents of which are incorporated herein by reference) may be used to prepare nanoparticles. In accordance with the present invention, single domain magnetically responsive nanoparticles are prepared by a

process comprising preparing a solution of magnetic metal salts and alkaline media to form a precipitate. The precipitate is then washed with a solvent like acetone and magnetically collected. The precipitate is washed again with the solvent and dried.

[0037] The mixture of magnetic metal salts may comprise an aqueous mixture of ferric chloride and ferrous chlorides in a ratio of between 2 to 1 and 10 to 1, which is added to the aqueous alkaline media. The alkaline media may comprise ammonium hydroxide. The combination of the magnetic metal salt mixture and the alkaline media results in a gelatinous precipitate that may be isolated from the solution by centrifugation or magnetic decantation without washing with water. The gelatinous precipitate may be peptized with, for example, Tetramethyl-ammonium hydroxide to form a stable alkaline magnetic solution or nanodispersion. Solutions of this type are stable for long periods of time. Acidic solutions can also be produced.

[0038] The resulting nanoparticles 12 can be collected from stable nanodispersion through the controlled reduction of pH to below 10.5 or less. At this point the nanoparticles 12 can be magnetically extracted and collected. The particles are easily dispersed again in aqueous media with sonication.

[0039] Because further processing of the nanoparticles to form nanospheres may be desired or required, it is not necessary to dry the nanoparticles at this stage, due to aggregation and agglomeration phenomena which may yield undesirable size distributions, and subsequent inefficient and ineffective performance properties. However, if the formation of nanospheres is desired, the nanoparticles may be either air dried or air dried and then oven dried.

[0040] If surface treatment of the nanoparticles is required, the precipitate may be surface treated with sodium silicate or chloride salts. At a high pH, a surfactant may be added and followed by the introduction of the coating material. As the pH is slowly reduced, the magnetic nanoparticles are coated with the silica.

[0041] Turning to Figure 6, there is shown therein, a system 50 for the preparation and production of magnetically responsive nanospheres having a bio-compatible shell. A feed stock comprising at least a magnetically responsive nanoparticle and a sodium silicate is prepared and atomized using the spray dryer system 50. It will be appreciated that the polymer and therapeutic may be added to the feed stock so that the resulting nanosphere contains a therapeutic. The nanodispersion feed stock 51 is introduced into the system 50 through a fluid inlet 52 and into a reservoir 54. The feed stock 51 is contained within the reservoir 54 until it is injected into a heated drying chamber 56 through a pressure spray nozzle 58. The spray nozzle 58 produces an aerosol distribution through ultrasonic liquid atomization. Evaporation of the solvent, diffusion of solute, drying of the nanoparticle, all occur inside the drying chamber 56 to form the nanospheres 10a, 10b, and 10c.

[0042] The composition of the nanospheres is determined by the solute or reactant concentrations in the starting nanodispersion solution, which is prepared in predetermined stoichiometric ratios. Water or alcohol may be used as a solvent, either separately or in combination. The colloidal suspension, which contains liquid and solid particles, is atomized into the drying chamber 56 and the liquid phase (the solvent) evaporates from the droplets.

[0043] The average size and size distribution of the final nanospheres may be roughly determined from the size of the atomized droplet and the initial concentration of the starting nanodispersion. The nanodispersion is forced out of the spray nozzle 58 by a compressed gas, for example, nitrogen. Atomization is the production of droplets and their dispersion into the gas, and the apparatus used to produce such droplets is known as an atomizer (not shown). The size or morphology of the final particles produced can also be determined by the concentration and velocity of the droplet generated by the atomizers. A variety of atomization methods may be used, such as air-assist (pneumatic) or a two-fluids nozzle, ultrasonic, vibrating orifice and spinning disk.

[0044] Various modifications of operating conditions in the spray dryer system 50 will lead to an efficient production of nanospheres of a desired particle size. Such modifications may include, for example, use of one or more atomizer nozzles, controlling the pressure at which the feed nanodispersion is pumped through the nozzle 58, and the feed to air ratio. Operating conditions, for example, the dispersion concentration, feed rate, nozzle concentration, gas pressure, and feed flow rate are specified to produce an aerosol distribution such that on drying, the resultant nanosphere will have a particle diameter of 100 nanometers or less.

[0045] The drying chamber 56 may optionally contain an electromagnetic coil 60 capable of generating a static or an oscillating magnetic field. As the atomized droplets pass through this applied magnetic field, the nanoparticles within the droplets are forced to align so that their magnetic moments are uniformly aligned. An operating value range for the magnitude of the magnetic field to be effective in causing the nanoparticles to be aligned may depend on, for example, the size of the nanoparticles or the size of the resultant nanosphere, and may be, in the range of .05 T to 10T. The alignment of the nanoparticles in the magnetic field during the drying process results in the production of magnetically responsive nanospheres having increased susceptibility. Such nanospheres with increased magnetic susceptibility will be easier to manipulate and vector in applications, responding more effectively in the magnetic field, which in turn may assist with site-specific positioning and internalization of the nanospheres.

[0046] It will be appreciated that cell adhesion factors may be added to the surface of the bio-compatible out shell by redispersing the nanospheres in a solution containing the desired adhesion factor. The solution may be aqueous, organic or a mixture of both. The above spray drying process is repeated using the spray drying system 50. This second spray drying provides a nanosphere having a bio-compatible outer shell that has adhesion factors showing an affinity for certain target cells.

[0047] In accordance with the present invention, there is provided a method for vectored delivery of nanospheres 10a and 10c, containing therapeutics to a desired site in a body. The method comprises using a three dimensional magnetic field to vector at least a nanosphere to the desired site within the body. It will be appreciated that a plurality of nanospheres may be used without departing from the spirit of the present invention.

[0048] The nanosphere 10a or 10c, containing the therapeutic, is introduced into the body by, for example, application of a paste containing the magnetically responsive nanosphere to the requisite body part to be treated. More specifically, where an organ to be treated is easily accessible, for example, an ear, the paste may be applied by any generally known method, for example, by a brush-type applicator. In the event that the organ to be treated is not readily accessible, the nanosphere 10a or 10c may be introduced close to the site with the use of other generally applicable methods, for example, a catheter.

[0049] The magnetically responsive nanospheres are three-dimensionally vectored toward the target site by the application of a three dimensional magnetic field to guide the nanosphere 10a or 10c the desired site where the nanospheres may be internalized by the cells. The three-dimensional magnetic field is created externally by, for example, an electromagnetic unit similar to the type used in rf-cardiac ablation surgery, of which the Stereotaxis Interventional Workstation is a known example.

[0050] In rf-catheter ablation surgery, utilization of an electromagnetic, three-dimensional, catheter Interventional Workstation aids the cardiac electrophysiologist in placing the recording/lesioning catheter. This technology integrates a super-cooled electromagnet which generates magnetic fields of about 0.2 Tesla to guide the tip of the ablation catheter to the target site in the heart, for example, to the right atrial appendage of the heart. The three dimensional magnetic field permits the catheter to enter and place its tip on difficult anatomical sites. However, because this unit creates a uniform magnetic filed, it is necessary to create a gradient

in the field in which nanospheres can be vectored towards the desired site. Once at the site, the nanospheres are held in place until internalized by cells has occurred. Internalization can generally be expected to occur within as much as a few hours or as little as a few minutes.

[0051] In yet another example, consistent with the embodiments of the present invention, the magnetically responsive nanospheres 10a and 10c may be used to treat urological diseases. In the event that there is a bacteria buildup, it becomes necessary to deliver drugs, such as antibiotics, to the infected region. However, traditional methods are not extremely effective due to the difficulty associated with the penetration of the antibiotics through the cell walls to the infected site. This is especially true in treatment of bacterial diseases that occur in human females. The magnetically responsive nanospheres overcome this difficulty due to the ease with which they are endocytosed and the ability to enhance internalization magnetically. Hence, therapeutic antibiotics transported with the nanosphere 10a or 10c may be delivered site-specifically. Cell internalization is facilitated by the use of a magnetic force, which is used to pull the nanoparticles through the cellular wall to the infection site. Additionally, adhesion factors may be used, as previously discussed, with the nanospheres to aid the process of endocytosis. The therapeutics may be delivered by, for example, a catheter or introduced through an injection at or near the infection site.

[0052] Consistent with the embodiments of the present invention, the nanospheres are vectored toward a target site based on gradients created in the magnetic field. The nanospheres, having superparamagnetic nanoparticles, when in a magnetic field are attracted to the strongest side of the gradient and will not be attracted to other or similar particles. Once the magnetic field is removed, the nanoparticles lose their magnetic properties, exhibiting little remanence.

[0053] In addition, or in the alternative, an external magnetic field from, for example, a permanent magnet positioned at an opposing end from where the nanoparticles are introduced

towards the cell, may be used to provide an external force to facilitate internalization into cells by drawing the nanoparticles into the cellular layer.

[0054] Once the nanospheres 10a or 10c have transported the therapeutic 16 to the desired site, the magnetic field may remain for a suitable length of time to allow the therapeutic to be internalized into the cells by the magnetic force. Residence time of the magnetic field depends on several factors, such as particle size and the applied external magnetic force.

[0055] It will be appreciated that the vectored therapeutic delivery system described herein can be used to deliver site-specifically a wide range of therapeutics including, but not limited to, chemotherapeutics for targeted cancer therapies, therapeutics for the treatment of gastric disorders such as Gastro-Intestinal-Reflux-Disease, and for therapeutics having a wide range of solubility properties -- soluble versus insoluble, thus, improving the effectiveness of the therapeutics while minimizing side effects.

[0056] The nanosphere 10b of Figure 2 may be magnetically vectored to a site so that the nanosphere may be incorporated into the cell structure of an organ for long-term assistance in organ functioning. In cases where mechanical function of an organ has failed or is diminished, magnetically responsive nanosphere 10b can be used in a corrective or remedial sense. Such nanospheres 10b may be used for various applications, such as, but not limited to, sphincter muscle opening and closing, blinking of an eye, tissue repair/reattachment, bladder control, ear vibration for sound amplification, and diagnostics such as imaging. The capacity to use magnetic organ assisting nanospheres 10b to assist in wound healing and tissue repair may improve healing rates and recovery times. Examples of such applications include connecting and holding torn ligaments and muscles during and after surgery; and controlling or stimulating involuntary muscle movements such as eye blinking. An exemplary embodiment may be a nanosphere having at least a magnetically responsive nanoparticle that is effective as a component of an implantable hearing device ("IHD").

[0057] The nanosphere 10b of Figure 2 is, in the IHD application, applied to the surface of the middle ear mucosal epithelium of the incus or the tympanic membrane and caused to be internalized, which can occur by absorption with the aid of cell adhesion factors 20 or by magnetically assisting by positioning a magnetic device on the opposite side of the organ, or a combination of the two.

[0058] Once internalized and placed in a magnetic field such as, but not limited to, that produced by an oscillating electromagnetic coil similar to that of existing IHDs, the embedded nanoparticles will vibrate, causing the ossicular chain to similarly vibrate, thus assisting the organs of the ear in amplifying sound.

[0059] In another application of the present invention, there are provided nanoparticles that are surface treated to render them useful as imaging tools. These surface treated nanoparticles may be prepared by any process or method discussed herein. The magnetically responsive nanoparticles may be surface treated with, for example, gold, gadolinium or titanium. Such surface treated nanoparticles may be vectored to a desired site with an external three dimensional magnetic field. The surface treated nanoparticles may provide a localized enhanced image. For example, gadolinium is a specific contrast agent used for detecting and highlighting neoplasia/inflammatory tissue for MRI evaluation, it is routinely utilized in most scan procedures. However, getting gadolinium to the site for accurate imaging has faced some difficulties that could be resolved through the use of 3-D vectoring of the nanoparticles as discussed above.

[0060] The invention will now be described in more detail with reference to the following Examples which merely serve to illustrate the invention, not to restrict or limit it in any way.

EXAMPLE 1

[0061] An aqueous solution of Ferric Chloride ($FeCl_3$) was mixed with an acidic solution of Ferrous Chloride (Fe_2Cl_3) in a molar ration of 2:1 to 10:1, and heated to 75°C – 100°C under an N_2 blanket, with gentle stirring, and held at that temperature for approximately 15 – 30 minutes. The Fe mixture was added to aqueous ammonia to form a magnetic-solution precipitate. The mixture was then stirred for 30 minutes under an N_2 blanket and the precipitate collected using a magnetic field. The precipitate was washed several times in distilled water to remove salt products produced by the reaction. The precipitate was collected using a magnetic field and dispersed in acetone, collected and dried two more times. The magnetically responsive nanoparticles produced by the above process had a magnetic susceptibility of greater than 35 – 40 emu/g and an average diameter of less than 50 nanometers.

EXAMPLE 2

[0062] The procedure according to Example 1 was followed to produce a known weight of nanoparticles. The nanoparticles were then dispersed in aqueous ammonia at pH >11 to form a stable ferrofluid. A known weight of sodium silicate was added to aqueous ammonia to give a desired molar ratio of Si:Fe between 0.5 and 10, and added to the prepared ferrofluid under a N_2 blanket and allowed to stir for 15 minutes. The pH was adjusted to 10.5 with HCl and the mixture was stirred an additional 2 hours. The pH was again adjusted to 9.0, and the mixture was stirred for 2 more hours.

[0063] To ensure complete silica coating of the nanoparticles, the pH was raised to 10.5 with stirring for 2 hours and then lowered to pH 9.0 with HCl. The product was collected using a magnetic field and washed in distilled water and acetone. The product was then collected and dried. The silica coated magnetically responsive nanoparticles produced in this manner had a magnetic susceptibility greater than 20 emu/g while having an average diameter of less than 50 nanometers. The silica coated nanoparticles had a composition ratio of of 0.5:1 to 5:1 Si to Fe.

EXAMPLE 3

[0064] A known weight of the silica coated nanoparticles produced in Example 2 were dispersed at room temperature in a small amount of distilled water to form a thick gelatinous mass. An amino-silane, such as 3-aminopropyltrimethoxysilane, was added to the aqueous mixture with stirring, and allowed to react under an N_2 blanket for 30 minutes. The product was recovered using a magnetic field and washed several times in distilled water. The product was taken up in distilled water and the pH was lowered the pH to 6.5 with HCl. The product was collected magnetically, washed in distilled water and dispersed and collected from acetone. The presence of the amine functionality was confirmed using the Kaisar test.

EXAMPLE 4

[0065] Magnetically responsive nanoparticles 12 were used to facilitate the vibration of the middle ear structure in an animal model. The lateral surface of the incus was coated with a suspension of nanoparticles 12 by placing 100 microliters of the nanoparticle suspension in physiological saline (pH of about 7.4) onto the lateral surface of the incus. At 8 and 15 days post-implantation, the animals were euthanitized and taken to a laser Doppler interferometry laboratory. An electromagnetic coil 7 mm in length, 2 mm in diameter was placed 2-3 mm from the incus and activated with sinusoidal voltage of 8-11 volts, at 1000 Hz. A reflective laser target 1 x 1 mm was placed on the incus, which was in tact with the malleus and stapes.

[0066] The external magnetic field vibrated the incus at 2000 Hz (due to the superparamagnetic property of the nanoparticles 12). The amplitude of vibration was small (~5 nm) but the principle of magnetically induced movement of the ossicular chain using magnetically responsive nanoparticles was demonstrated. In two other animals these same nanoparticles 12 were placed on the tympanic membrane "TM" and an external magnetic field used to facilitate internalization of the nanoparticles into the epithelium. When the same electromagnetic coil was placed 2-3 mm from the TM and activated at 1000 Hz, 11 volts, it

vibrated at 2000 Hz with displacement amplitude of 16.5 nm. Thus, nanoparticles generated forces in the middle ear, thereby, aiding hearing amplification.

[0067] Various modifications can be made in the design and operation of the present invention without departing from the spirit thereof. Thus, while the principal preferred construction and modes of operation of the invention have been explained in what is now considered to represent its best embodiments, which have been illustrated and described, it should be understood that within the scope of the appended claims, the invention may be practiced otherwise than as specifically illustrated and described.